

### Sample Prelab for Module 3

There are two types of synapses in the brain, electrical and chemical synapses. In this lab, we will study chemical synapses by examining excitatory post synaptic potentials which are caused by the opening of ion channels. The transmission of information at a chemical synapse involves the conversion of an electrical signal in the presynaptic neuron to a chemical signal and then back to an electrical signal in the postsynaptic neuron. Depolarization of the presynaptic neuron causes an influx of calcium at the nerve terminal, therefore allowing the release of synaptic vesicles. The transmitters in the vesicles then diffuse across the synaptic cleft and bind to receptors on the postsynaptic neuron. The binding of glutamate to AMPA receptors on the postsynaptic neuron is responsible for most of the fast excitatory synaptic transmission in the central nervous system.

In this lab, our goals are to observe EPSPs at glutamatergic synapses from the Schaffer collaterals onto neurons in the CA1 region. To do this, we would have to insert a stimulator into the Schaffer collateral region while patch clamping a neuron in the CA1 region at the same time. Next, set capacitance compensation and bridge balance and then turn on the stimulator. To activate the neurons, inject 100 $\mu$ s shocks while adjusting the stimulus current until the initiation of EPSPs of 3-5mV and then get 15 sweeps of data while waiting 10 seconds between each sweep. Moving on to the next part of experiment 1, change the resting membrane potential to -80 mV and repeat part 1. Finally, move the resting membrane potential to -60mV and repeat part 1 again. For the second part of the experiment, paired synaptic stimuli are delivered at 4 different interstimulus intervals to elicit EPSPs of 3-5 mV. Once again, we will record 15 sweeps for each interval while waiting 10 seconds between each sweep. For the last experiment, we are going to change the solution bathing the brain slice from normal saline to a solution containing twice as much calcium as the normal solution. We will repeat experiment 2 with the brain slice bathed in the high calcium concentration solution.

The purpose of the first experiment is to see whether changing the resting membrane potential would affect the amplitude and shape of the EPSP. The purpose of the second experiment is to observe paired pulse facilitation by delivering paired pulse stimuli. Lastly, the purpose of the third experiment is to examine the effect of calcium in the presynaptic terminal by bathing the brain slice in a solution containing a higher concentration of calcium.

In the first experiment, the independent variable is the resting membrane potential and the dependent variable is the voltage. My hypothesis is that the lower the resting membrane potential, the greater the EPSP. For the second experiment, the independent variable is the interstimulus interval and the dependent variable is the voltage. My hypothesis is that the smaller the interstimulus interval, the larger the second EPSP of each paired pulse. For the last experiment, the independent variable is the solution bathing the brain slice and the dependent variable will once again be the voltage. My hypothesis is that the EPSPs elicited when the brain slice is in the high calcium concentration solution will be larger than the EPSPs when in normal solution.

I made my prediction about the first experiment based on the knowledge that at a lower resting membrane potential, there's a greater driving force for sodium to come in through AMPA receptors and this will cause a larger EPSP. This is because the equilibrium potential for sodium is about 60 mV and around 0 mV for AMPA receptors. Therefore, when the membrane is hyperpolarized, it's moving further away from the equilibrium potential for sodium and thus causes a greater driving force for sodium to go into the cell. I arrived at my hypothesis for the second experiment based on the fact that at shorter stimulus intervals, there can be a residual amount of calcium from the first stimulus that can affect the amount of calcium in the presynaptic neuron. This larger amount of calcium will allow more transmitters to be released during the second stimulus and create a greater EPSP. For the last experiment, I based my

hypothesis on the fact that at a higher external concentration of calcium, there's a greater driving force for calcium to flow into the cell so more vesicles will be released and thus, the EPSPs will be larger than when the brain slice is in normal saline.